IJCRT.ORG ISSN : 2320-2882

INTERNATIONAL JOURNAL OF CREATIVE RESEARCH THOUGHTS (IJCRT)

An International Open Access, Peer-reviewed, Refereed Journal

Exploring Soursop's Potential As An Insecticide Against Whiteflies

[1]Bibin VA, [2]Akshaya Rajan P, [3]Asna Haseen, [4]Vrundamohan, [5]Anu Jagajith A, [6]Dr. Lal Prasanth M L

[1]Eighth semester B. pharm Dr. Moopen's college of pharmacy, [2]Eighth semester B. pharm Dr. Moopen's college of pharmacy, [3]Eighth semester B. pharm Dr. Moopen's college of pharmacy, [4]Eighth semester B. pharm Dr. Moopen's college of pharmacy, [1]Associate professor of Dr. Moopen's college of pharmacy, [1]Principal of Dr. Moopen's college of pharmacy.

[1]Department of pharmacognosy,

[1]Dr. Moopen's college of pharmacy

Abstract: This work investigates the potential effect of soursop extracts as a natural insecticide against whiteflies. Utilizing various extraction techniques, active compounds are isolated from the leaf of the soursop plant and subjected to characterization study by using HP LC-MS and the active constituents found as 2-Hexynoic acid, acetic acid, 2-isopropoxyethylpropionate, 1,5-anhydro-d-mannitol then 2-hexynoic acid possess insecticidal activity. Whiteflies (*Hemiptera:Aleyrodidae)* are agricultural pest causing significant damage to a wide range of crops in world wide. So effective and environment friendly insecticides to manage whitefly populations have greater importance. Soursop is a tropical plant known for its medicinal properties, has botanical insecticide due to its bioactive compounds. The findings suggest that soursop extracts possess insecticidal properties against whiteflies, exhibiting repellent effects and causing mortality in treated insects. This study offers about using soursop as a substitute for synthetic insecticides in the control of whiteflies which offers an eco- friendly and natural pest control.

Index Terms **– Soursop, Whiteflies, Soxhlet extraction, HP LC-MS**

I. INTRODUCTION

Insect

Insects are incredibly diverse creatures, with over a million species identified. They play vital role in ecosystems as pollinators, decomposers, and prey. But some species can also be pests, causing damage to crops and transmitting diseases. Insects belongs to the largest class of the phylum Arthropoda, it's the most diverse group of organisms on the earth. Many of the insects are beneficial, some species are harmful, causing damage to crops, spreading diseases, or being household pests.

Whiteflies

Whiteflies are small insects of the family Aleyrodidae well- known for the great damage they can cause to a myriad of plant species. As they are sap suckers, they feed on the underside of the leaves by piercing their thin and delicate mouthparts, these winged pests are the smallest kind and closely related to aphids and scales. Especially harmful to greenhouses, and gardens, and agricultural crops, whitefly is a major cause of concern in both ornamental and other plants with its host.

Fig 1. Whiteflies

Several factors have led to the high incidence of whitefly infestations. Temperature and high humidity can create optimum conditions for whitefly populations to burst and so greenhouses are particularly susceptible. It also allows pests to multiply quickly as controlled environment such as greenhouses have no natural predators. However, the movement of plants carrying whitefly-transmitted gemini viruses accelerates their spread, both to new areas and to new hosts In addition, the repeated use of chemicals for controlling whiteflies may also stimulate the development of insect resistance to chemicals and also the reduction in populations of insects that is directly competing for the whitefly adults.

There are numerous species of whiteflies that are frequently seen; each has unique traits and favoured host plants. The Silverleaf Whitefly, or Bemisia tabaci, is well-known for its wide host range, which includes field crops, ornamentals, and vegetables. A common pest in greenhouses, the Trialeurodes vaporariorum, or Greenhouse Whitefly, harms a wide range of food and ornamental plants. Citrus trees are the primary target of the Dialeurodes citri, also known as the Citrus Whitefly, whereas the Aleurothrixus floccosus, also known as the Woolly Whitefly, is found on citrus and other fruit trees and is identified by the waxy filaments it creates.Whiteflies harm plants and the larger ecosystem in a variety of ways. Whiteflies ability to suck plant sap causes yellowing, wilting, stunted development, and leaf drop, therefore direct feeding harm is the main cause for worry. In addition, whiteflies secrete a sticky material known as honeydew, which promotes the development of sooty Mold on foliage. This Mold has the ability to hinder photosynthesis, worsening the plant's condition. More concerningly, whiteflies are vectors for a number of plant viruses, such as the Cucumber Mosaic Virus (CMV) and the Tomato Yellow Leaf Curl Virus (TYLCV), which can cause large output losses in crops impacted. Because of lower yields and higher control expenses, heavy infestations in agricultural contexts can result in significant financial losses. Furthermore, overusing chemical pesticides to suppress whitefly populations can upset the ecosystem's delicate balance by eliminating helpful insects and resulting in new pest outbreaks.

Whitefly infestations must be managed effectively with an integrated strategy incorporating chemical, biological, and cultural techniques. Removing affected plants, applying reflective mulches, and utilizing sticky traps to lower whitefly numbers are examples of cultural management. Biological measures aim to manage the population of whiteflies by introducing natural predators such parasitic wasps, lacewings, and lady beetles. Chemical controls ought to be applied sparingly even though they are occasionally required. Neem oil, insecticidal soaps, and selective insecticides can all be useful, but caution must be exercised to prevent resistance growth and damage to beneficial insects.

In summary, minimizing the effects of whiteflies on plants and the environment requires an awareness of the causes of infestations, the recognition of common species, and the recognition of the harm they do. Utilizing a blend of cultural, biological, and chemical management techniques, whitefly populations can be efficiently regulated to save crop and ornamental plants from their detrimental impacts.

Biopesticides

Biopesticides are certain types of pesticides derived from such natural materials as animals, plants, bacteria, and certain minerals. For example, canola oil and baking soda have pesticidal applications and are considered biopesticides. Also, there are fungi that control certain weeds, and other fungi that kill specific insects.

Types of biopecticides

Biopesticides are classified as follows:

- Microbial Pesticides:

  Microbial pesticides consist of a microorganism (eg: a bacterium, fungus, virus, or protozoan) as the active ingredient.

  Microbial pesticides can control many different kinds of pests, although each separate active ingredient is relatively specific for its target pest.

  Bacterial pathogens used for insect control are spore forming, rod shaped bacteria in the genus Bacillus which occur in soils.

  The most widely used microbial pesticides are subspecies and strains of Bacillus thuringiensis (Bt.). Each strain of this bacterium produces a different mix of proteins, and specifically kills one or a few related species of insect larvae. While some Bts control moth larvae found on plants, other Bts are specific for larvae of flies and mosquitoes.

  The target insect species are determined by whether the particular Bt produces a protein that can bind to a larval gut receptor causing the insect larvae to starve.

- Plant-Incorporated Protectants (PIPs)

  Plant-Incorporated Protectants (PIPs) are pesticidal substances that plants produce from the genetic material that has been added to the plant. For eg: Scientists can take the gene for Bt pesticidal protein and introduce the gene into plant's own genetic material. Then the plant instead of Bt bacterium manufactures the substance that destroys the pest.

  These are used to reduce destruction of crops by arthropod pests.

  Include genetically modified plants to express genes encoding insecticidal toxins.

- Biochemical pesticides:

  These are naturally occurring substances that control pests by nontoxic mechanisms.

  Biochemical pesticides include pheromones, plant extracts and natural insect growth regulators.

  Include substances such as insect sex pheromones that interfere with the mating and various scented plant extracts that attract pests to traps and natural insect growth regulators.

Advantages

- Less toxic than conventional pesticides.
- Affect only target pest and closely related organisms.
- Effective in very small quantities
- Decompose quickly resulting in lower exposures and largely avoiding pollution problems caused by conventional pesticides

Disadvantages

- A slower rate of control and often a lower efficacy and shorter persistence compared to conventional pesticides.
- Greater susceptibility to adverse environmental conditions.

- Greater level of knowledge required by the grower to use them effectively.

Biopesticide uses in india

For the past 10 years, India has witnessed a tremendous increase in the use of biopesticides as a significant crop protection technique. Mainly as the status shows, production techniques are standardized, are like Trichoderma, Gliocladium, Paecilomyces, Pseudomonas, Trichogramma, NPV, and bacillus are used to counter many insects and pest problems. It's been see many times that biocontrol agents are used successfully in India. Some examples are as follows;

• Telonemia scrupulous- it is a bug that has been used to control the tremendous increase of lantana weed in the fields.

• Epiricania melanoleuca and Tetrasticus pyrillae- many states suffered from a situation of sugarcane perilla, which was successfully controlled by using these natural enemies of pyrilla.

• Trichogramma – an organism that feeds on the eggs of sugarcane borers and used to counter borers in many states, mainly Tamil Nadu, Rajasthan, UP, Bihar, and Haryana. Trichogramma has also been used against rice stem boresrs and lead folders.

• Bracon, Chelonus and Chrysopa spp- these are used to counterattack cotton bollworms.

• Coccinellid- these are a species of beetles that are used against sugarcane scale insects since these are a predator of those insects. This has resolved sugarcane yield loss in major states such as UP, West Bengal, Gujarat, and Karnataka.

Plant

The soursop plant scientifically known as Annona muricata L. is a tropical evergreen tree belonging to the Annonaceae family. Tropical plant species A. muricata L. is well-known for its edible fruit. The plant or part of the plants that are used for various purposes. It consists of flower, fruit, bark, leaf, root, seeds. A. muricata L. commonly called soursop, is part of the Annonaceae family, which comprises more than 130 genera and 2300 species. The soursop has some medicinal uses. A decoction of young shoots and leaves is a remedy for gall bladder infection, coughs, diarrhea, dysentery and indigestion. Mashed leaves are used for treatments of eczema and rheumatism. The flowers are antispasmodic. The ripe fruit prevents scurvy while the unripe fruit may be used for dysentery and has astringent properties.
They are widely grown in tropical and subtropical areas, such as Southeast Asia, South America. Soursop is the most tropical of the Annona species and is considered a plant of low altitudes and a hot and humid climate. It is grown primarily at altitudes lower than 900 m above sea level. However, good productive orchards are found at altitudes of up to 1100 m. Soursop adapts well to tropical humid regions all over the world. Soursop grows and produces well at 21 to 30ºC.It is very sensitive to severe changes in temperature, especially if it goes below 12ºC. In mountainous areas where temperature ranges between 15°-25° C, the tree produces very few fruit. Frost kills, young trees but older trees show some tolerance. Very low temperature, higher temperature and low humidity are detrimental for pollination and fruit formation. Moderate temperature (25-30° C) and high humidity (80 %) greatly improves pollination and fruiting. Soursop can be is grown on many soil types but sandy to sandy loam soils of medium texture are most suitable. Soil pH should be between 6.0 and 6.5. The soil should be rich in organic matter and low in salts. Best growth can be achieved in deep, rich, well-drained, semi-dry soils. Good drainage is essential to minimize the incidence of collar rot. Water logging is a major cause of floral abscission and root rot. Sour sop cannot tolerate water stagnation but can tolerate dry soil conditions. The Water logged and soil without drainage should be avoided for soursop cultivation. Fertilizer should be applied around the plant but only lightly incorporated into the soil to avoid damaging the developing root system. The fruit is picked when full grown and still firm but slightly yellow-green.
The plant which produces edible fruit round a year and is widely used and it is widely used as a traditional medicine for skin disease, respiratory disease, fever, bacterial infections, diabetes, hypertension, and cancer. Different parts of A. muricata L. which shows different activities. Phytochemically, they contain constituents

are acetogenin, alkaloids, and flavonoids. leaf extract revealed secondary metabolites such as terpenoids, saponins, coumarins, lactones, anthraquinones, glycosides, tannins, and phytosterols.

Mode of action that these acetogenins are super inhibitors of enzyme processes that are only found in the membranes of cancerous tumor cells. Acetogenins are characterized by a long aliphatic chain of 35 to 38 carbons bonded to a c-lactone ring, terminally substituted by b unsaturated methyl (sometimes it is a Ketolactone), with one or two Tetrahydrofurans (THF) located along the hydrocarbon chain and a determined number of oxygen groups (hydroxyl, acetoxyls, ketones, epoxy). Most of the acetogenins found in A.muricata L. contain a THF ring, although acetogenins have also been reported with two adjacent or nonadjacent THF rings. Acetogenins are linear and may have one or two epoxy groups. The isolate acetogenin from Annona muricata L. by thin layer chromatography and column chromatography by using kedde's reagent.

II. MATERIALS AND METHODS

Collection and authentication of plant material

The leaves of A. muricata L. were collected from the various areas of the Wayanad district. The leaf specimen is authentified by Dr. Raji R, Associate professor, Department of Botony, St. Mary's college, Sulthan bathery, Wayanad.

Fig 2. Plant *A. muricata L*

Synonyms: Guanabana, soursop, graviola, Brazilian paw paw.

Vernacular names

Table 1. Vernacular names of the plant

Language	Names
English	Soursop
Malayalam	Mullanchakka, cancer chakka, mullatha
Hindi	Khatta
Sankrit	Lavaniyam

Geographical distribution

It is grown mostly in tropical region of the world from South America to Australia, Asia and Africa.

Plant taxonomy

Table 2. Plant taxonomy

Kingdom	Plantae
Division	Spermatophyta
Subdivision	Angiospermae
Class	Dicotyledonae
Order	Polycarpiceae
Family	Annonaceae
Genus	*Annona*
Species	*muricata L*

Method of propogation

The plant is propogated through seeds and stem cuttings.

Medicinal properties

A. muricata L. which having several medicinal properties such as antiulcer, antimalarial, Antidiarrhea, Antidiabetic, Antiprotozoal, Antidiabetic, Antibacterial, Antiviral, Antihypertensive, Wound Healing, anticancer etc.

Macroscopical description

Leaves of *A. muricata L.* were subjected to macroscopical evaluation for parameters like colour, size, taste, shape by using simple microscope.

Microscopic description

Transverse section of leaves

The microscopic evaluation leaves were sectioned in transverse direction. The plant leaves are cross section was prepared and it is stained by the use of HCL and phloroglucinol. Viewed at 40x and 10x.

Powder microscopy

For performing the powder microscopy, the powder of leaves was taken and stained using HCL and phloroglucinol and then it is mounted on a glass slide and viewed under microscope. viewed at 40x.

Moisture content determination

A tarred evaporating dish was heated to a constant weight and stored in a desiccators, 2g of the powdered plant material was added to the dish and kept in an oven maintained at a temperature of 105 0c. It was allowed to dry until a constant weight was achieved. The difference in weight of the evaporating dish was noted.

Ash value

A tarred nickel crucible was ignited to a constant weight at a dull red heat, cooled and stored in a desiccator. A 2g of the powdered material was weighed into the nickel crucible and heated gently until all the moisture had been driven off and the material had been completely charred. The heat was increased until most of the carbon had been completely vaporized after which the material was heated to about 450 oc to make the residue carbon free. The residue was cooled and weighed. The heating and cooling were continued until a constant weight was achieved.

Extraction of *A. muricata L. leaves*

The extraction of leaves is done by using Soxhlet method. The leaves were collected and it is shade dried for 4-5 days. After proper drying the leaves were powdered.

Soxhlet method

In the Soxhlet method, about 60g of powdered drug was taken and it is packed into thimble. 1L of ethanol (95%) was taken and added to round bottom flask which is connected to the Soxhlet extractor. 24hrs of extraction was performed.

Sample preparation

After the extraction, the extract was evaporated and then subjected to drying. After drying the dried material was redissolved in acetone with the aid of charcoal to remove the impurities and filtered. The resulting mixture was dried and powdered, and placed in a Buchner funnel over a layer of silica gel 60.

Identification of active constituents by TLC

TLC was carried out for the identification of active constituents present in the leaf extract of plant A. muricata L. TLC plate is prepared by using silica gel 60 and then spot is identified by using UV spectrophotometer.

$$\text{Rf value} = \frac{\text{Distance travelled by the solute}}{\text{Distance travelled by the solvent}}$$

Isolation of active constituents by column chromatography

After the extraction, the extract was evaporated and then subjected to drying. The dried material was redissolved in acetone and filtered using charcoal. The resulting mixture was dried and powdered, and placed in a Buchner funnel over a layer of silica gel 60 (230–400 mesh). Elution was performed stepwise with each of water, ethanol, ethanol ethyl acetate, chloroform. The fractions were combined on the basis of their similarity into five groups: F1 – water, F2 – water: ethanol, F3 – water: Ethanol (1:1), F4 – Ethanol: Ethyl acetate (1:1) and F5 – chloroform. The whole of F4 was submitted to CC over silica gel 60 and fractions were evaporated. This product was subjected to and also analysed by TLC and LC-MS.

Phytochemical screening

Different qualitative tests for identification of phytoconstituents conducted.

Phytochemical analysis of extract by LC MS

LC MS extract phytochemical analysis was studied by using the LC-MS at Centre for biotechnology and phytopharmacognosy research, Coimbatore. The chemical constituents of the ethanolic extracts were determined using LC-MS. LC-MS analysis was performed using Mariner Bio spectrometry equipped with a binary pump. The HPLC was interfaced with a Q-TOF mass spectrometer fitted with an ESI source. Full-scan mode from m/z 100 to 1200 was performed with a source temperature of 140°C. HPLC column Phenomenex 5µ C8, (150 × 2 mm.) was used for the analysis. Solvent was acetonitrile with 0.1% formic acid in H2O. Column: X-Bridge C18 (150*4.6) 3.5µm. Solvents were delivered at a total flow rate of 0.1 mL/min. The solvent was run by isocratic elution. The MS spectra were acquired in the positive ion mode.

Biological activity

In vitro insecticidal activity

To carry out insecticidal activity of plant, the insects were collected in a container. The stock solution 50ml was made by introducing 5g of dried sample of A. muricata L. into distilled water and different concentration of extract were prepared as 2mg/ml, 4mg/ml and 6mg/ml and 1 considered as control group. Mortality of insects was observed for 25 minutes. When a needle was used to probe the insect if they did not respond it was decided that they were dead.

$$\text{Percentage mortality} = \frac{\text{Number of dead insect}}{\text{Number of insects introduced}} \times 100$$

III. RESULTS AND DISCUSSION

Macroscopic description

In macroscopic description the plant material is classified based on sensory evaluation like color, odour, shape etc.

Leaves

The plant that produces usually has large, glossy leaves that are dark green in color and it have entire margin, oblong or ovate shapes, symmetrical base, acute apex, and length of 7 to 20cm.

Fig:2.1 leaf of *A. muricata L.*

Flower

They are generally small, with a diameter ranging from about 2 to 3 cm. Soursop flowers have a pale green to yellowish-green color and consist of three outer, fleshy, triangular petals and three inner, smaller, more narrow petals. Flowers are hermaphrodite, usually fragrant, and solitary or in groups of two or four, with fascicles of three green sepals and six petals arranged in two verticals.

Fig.2.2. Flower

Fruit

The fruit which is large and green fruit with a spiky, irregular surface & size about 8cm to 12cm length. Dark green color on fruit which indicates the fully matured and ripened fruit.

Fig.2.3. Fruit

Seeds

The seeds are dark brown to black in color and are 2 to 3 cm in length. It contains smooth, shiny surface and are oblong or oval-shaped. Seeds are surrounded by a white, fibrous pulp within the fruit.

Fig.2.4. Seeds

Microscopic characters

Transverse section of leaves

The transverse section of leaf A. muricata L. shows cuticle, hypodermis, trichomes, phloem fibers, xylem vessels, lower epidermis, lower cuticle.

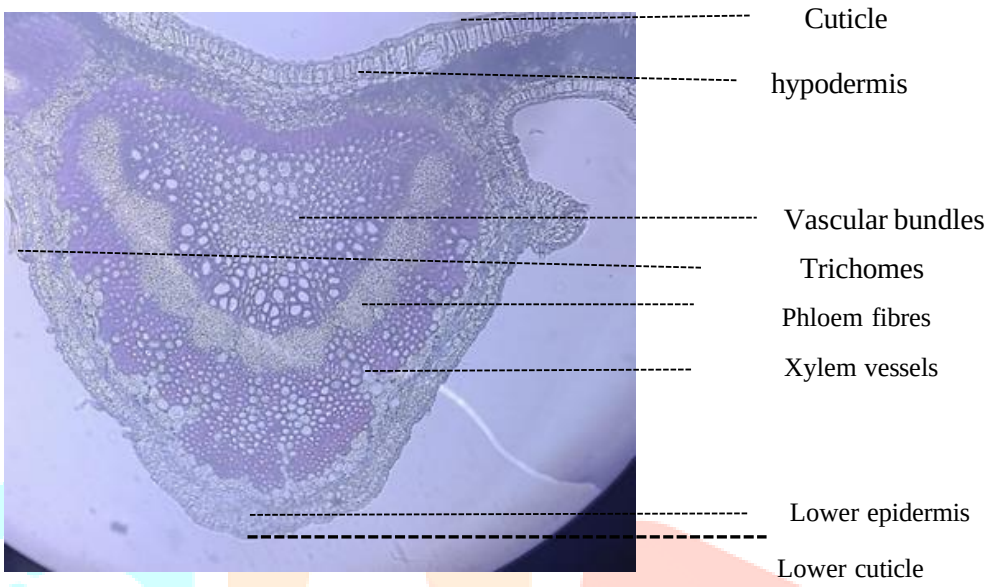
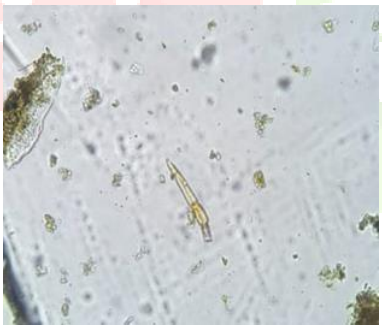
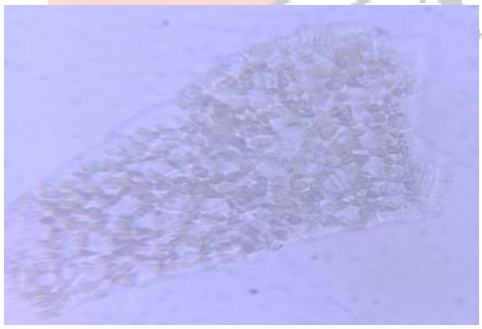


Fig.2.5. T.S of Plant

Powder microscopy

The powder characters of A. muricata L. shows trichomes, xylem vessels, calcium oxalate crystals, epidermis.





Moisture content

The moisture content of the given powdered material was 6.50%w/w.

Ash value

The total ash value of powdered material was found to be 10.90.

Extraction of *A. muricata L.* leaves

Fig.3. Soxhlet extraction

Identification of active constituents by TLC

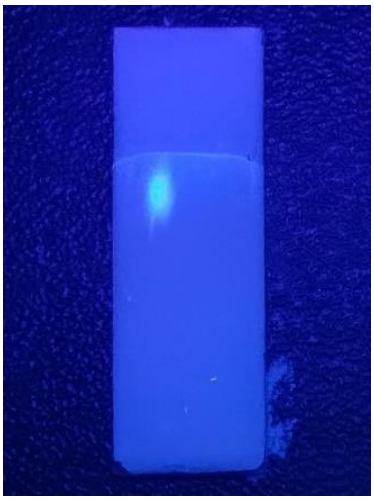


Fig.4. TLC Plate

Rf value was found to be = 0.41.

Phytochemical screening

The table shows the phytochemicals present in the extract of leaves of *A. muricata L.* leaves.

SL No	CONSTITUENTS	RESULT
1	Alkaloids	+++
2	Tannins	+++
3	Flavonoids	+
4	Carbohydrates	++
5	Glycosides	+

Table.3.Phytochemical screening

Characterization of extract by LC-MS

The characterization of LC-MS extract via LC-MS analysis are presented. The compounds were detected and were identified by their fragmentation pattern. Peak area and retention time were used for the identification of the compounds.

S. NO	Phytochemicals	m/z
1	Acetic acid, [(aminocarbonyl)amino] oxo-	132.92
2	2-Hexynoicacid	112.19
3	2-Isopropoxyethylpropionate	160.98
4	1,5-Anhydro-d-mannitol	165.20

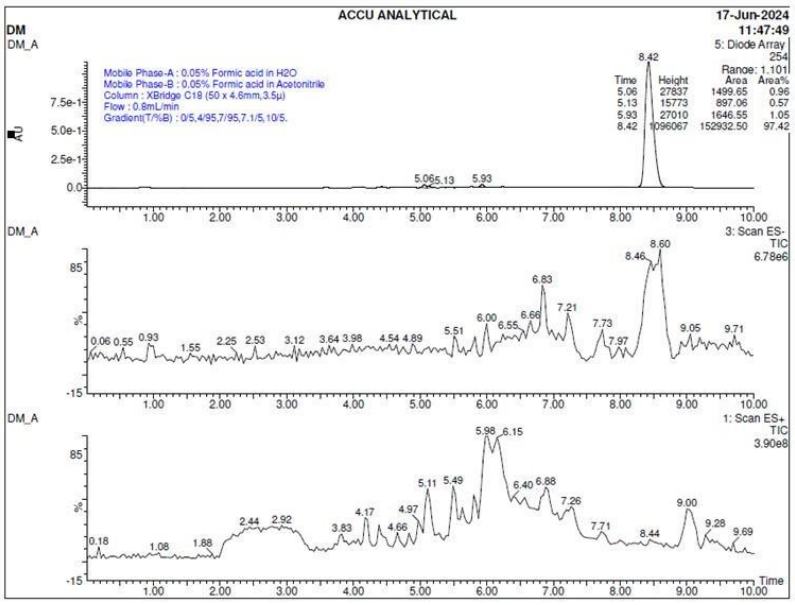


Fig.6. Graphical representation of phytoconstituents in LCMS

Biological activity

The study was conducted to evaluate the insecticide activity of soursop plant against white flies. 10 white flies are introduced in each petri dish contain different concentrations of plant extract as 2mg/ml, 4mg/ml, 6mg/ml and water is considered as control group. Mortality was observed for 25 minutes.

SL. No.	Concentrations	Mean mortality	Percentage mortality
1	2mg/ml	2.67	26.67
2	4mg/ml	4.67	46.67
3	6mg/ml	8.33	83.33
4	Control group	3.33	33.3

Table.4. The mean mortality and percentage mortality

The plant was found to have concentration dependent activity against white flies, the percentage mortality increased with increasing concentration of the plant extract. The result shows that 6mg/ml concentration of A.muricata L. leaf extract have the highest percentage of mortality among the various plant extract concentrations.

Fig.6. Insecticidal activity of plant extract

Statistical analysis

The data is collected and entered in Ms. Excel analysis was done using SPSS 26.0 version. The variables are expressed in Mean, Standard deviation, Minimum, maximum etc. To compare between different concentration ANOVA is used.

Descriptive statistics		Value for 25 minutes
N		3
Mean		4.75
Std. Deviation		2.42
Minimum		13
Maximum		18
Percentile	**Q1**	25
	Median	50
	Q3	75

Table.5. Descriptive statistics

Concentrations	Mean ±SD	95% of confidence interval for mean		P value
2mg/ml	2.67 ± 0.58	1.23	4.10	
4mg/ml	4.67 ± 1.53	0.87	8.46	**<0.001**
6mg/ml	8.33 ± 0.58	6.90	9.77	
Control group	3.33± 0.58	1.90	4.77	
Total	4.75 ± 2.42	3.21	6.29	

Table.6. ANOVA

Graphical representation of insecticidal effect of plant extract

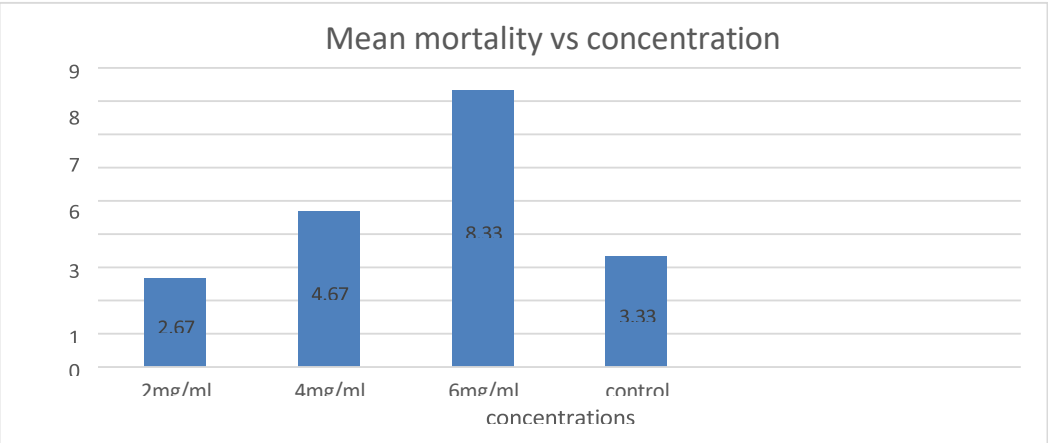


Fig.7. Graphical representation of insecticidal effect of plant extract

SUMMARY & CONCLUSION

The objective of the presented work was to evaluate insecticidal activity of A. muricata L plant extract against white flies was done. Initially we performed the literature review, microscopic and macroscopic characters including colour, size, powder microscopy, transverse section, ash value moisture content etc. The extraction of Annona muricata L. was carried out by Soxhlet extraction by using ethanol as solvent. Phytochemical studies show the presence of alkaloids, carbohydrates, tannins, saponin, flavonoids. The column chromatography and TLC were performed. The fraction from the column chromatography was subjected to LC- MS for the detection of active constituent present in the extract. The following compounds were detected and were identified by their fragmentation pattern.

1.Acetic acid, [(aminocarbonyl)amino] oxo-
2.2-Hexynoicacid
3.2-Isopropoxyethylpropionate
4.1,5-Anhydro-d-mannitol

From these compounds the 2-Hexynoicacid has been reported to exhibit insecticidal activity against whiteflies. The insecticidal activity of 2-Hexynoicacid against whiteflies was found to be concentration dependent, i.e the fraction obtained from column chromatography was prepared in to different concentrations such as 2mg/ml, 4mg/ml, 6mg/ml and subjected to insecticidal activity. Water is considered as control group. Among the three concentrations, 6µg/ml shows the higher mortality rate when compared to other concentrations. The higher concentration resulting higher mortality rates. 2-Hexynoicacid also exhibited repellent activity against whiteflies, with a significant reduction in whitefly settling and feeding behavior observed at higher concentrations.

Soursop which offers a natural alternative to synthetic insecticides, potentially reducing the environmental and health impacts associated with chemical pesticides. Soursop based insecticides could be used in organic farming, reducing the reliance on synthetic pesticides and promoting eco- friendly practices and also it provide opportunities for growers and communities involved in soursop cultivation.

REFERENCES

1. Stern VM, Smith RF, Van den Bosch R, Hagen KS. The integrated control concepts. Hilgardia.1959;29(2);81-101.
2. Pedigo LP, Rice ME. Entomology and pest management. 6th ed. Upper Saddle River: Prentice Hall;2009.
3. Kogan M. Integrated pest management: Historical perspectives and contemporary developments. Annu Rev Entomol.1998;43:243-70.
4. Oerke EC. Crop losses to pests. J Agric Sci. 2006;144(1):31-43.
5. Brown JK, Frohlich DR, Rosell RC. The sweet potato or silverleaf whiteflies: biotypes of Bemisia tabaci or a species complex. Annu Rev Entomol.1995;40:511-34.
6. Byrne DN, Bellows TS. Whitefly biology. Annu Rev Entomol.1991;36:431-57.
7. Liu TX, Stansly PA. Insecticidal control and resistance management for Bemisia tabaci. Crop prot. 1995;14(4):255-63.
8. De Barro PJ, Liu SS, Boykin LM, Dinsdale AP, Bemisia tabaci: A statement or species status. Annu Rev Entomol.2011;56:1-19.
9. Sudakin DL. Biopesticides. Toxicological reviews. 2003 Jun;22:83-90.
10. Seiber JN, Coats J, Duke SO, Gross AD. Biopesticides: state of the art and future opportunities.

Journal of agricultural and food chemistry. 2014 Dec 3;62(48):11613-9.

11. Gupta S, Dikshit AK. Biopesticides: An ecofriendly approach for pest control. Journal of Biopesticides. 2010;3(Special Issue):186.
12. Kumar KK, Sridhar J, Murali-Baskaran RK, Senthil-Nathan S, Kaushal P, Dara SK, Arthurs S. Microbial biopesticides for insect pest management in India: Current status and future prospects. Journal of invertebrate pathology. 2019 Jul 1;165:74-81.
13. Yadav R. Biopesticides: Current status and future prospects. Proceedings of the International Academy of Ecology and Environmental Sciences. 2022 Sep 1;12(3):211.
14. Tripathi YN, Divyanshu K, Kumar S, Jaiswal LK, Khan A, Birla H, Gupta A, Singh SP, Upadhyay RS. Biopesticides: current status and future prospects in India. Bioeconomy for sustainable development. 2020:79-109.
15. Kumar S. Biopesticides: a need for food and environmental safety. J Biofertil Biopestic. 2012;3(4):1-3.
16. Dar SA, Khan ZH, Khan AA, Ahmad SB. Biopesticides–Its Prospects and Limitations: An Overview. Perspective in Animal Ecology and Reproduction. New Delhi, India: Astral International (P) Ltd. 2019:296-314.
17. Parmar BS. Biopesticides: an Indian overview. Pesticide Research Journal. 2010;22(2):93-110.
18. Aneja KR, Khan SA, Aneja A. Biopesticides an eco-friendly pest management approach in agriculture: status and prospects. Kavaka. 2016;47: 145-54.
19. Idowu OP, Olubunmi SO, Peter A. The Biology and Ecology of Annona muricata l., Soursop; State of Knowledge. Chemical and Biomolecular Engineering. 2022 Oct 17;7(3):46.
20. Patel MS, Patel JK. A review on a miracle fruits of Annona muricata. Journal of Pharmacognosy and Phytochemistry. 2016;5(1):137-48
21. Badrie N, Schauss AG. Soursop (Annona muricata L.): composition, nutritional value, medicinal uses, and toxicology. InBioactive foods in promoting health 2010 Jan 1 (pp. 621-643). Academic Press.
22. Terán-Erazo B, Alia-Tejacal I, Balois-Morales R, Juárez-Lopez P, López-Guzmán GG, Pérez-Arias GA, Núñez-Colín CA. Physical, chemical and morphological fruit characterization of soursoup (*Annona muricata L.*). Agrociencia (Montecillo). 2019;53(7):1013-27.
23. Gleye, C.; Laurens, A.; Hocquemiller, R.; Cavé, A.; Laprévote, O.; Serani, L. Isolation of montecristin, a key metabolite in biogenesis of acetogenins from *Annona muricata* and its structure elucidation by using tandem mass spectrometry. *J. Org. Chem.* **1997**, *62*,
24. Mutakin M, Fauziati R, Fadhilah FN, Zuhrotun A, Amalia R, Hadisaputri YE. Pharmacological activities of soursop (Annona muricata Lin.). Molecules. 2022 Feb 10;27(4):1201.
25. Qorina F, Arsianti A, Fithrotunnisa Q, Tejaputri N, Azizah NN, Putrianingsih R. Cytotoxicity of soursop leaves (Annona muricata) against cervical HeLa cancer cells. Pharmacognosy Journal. 2020;12(1).
26. Dash GK, Hashim MH, Hassan AK, Muthukumarasamy R. Pharmacognostic studies on the

leaves of *Annona muricata Linn.*

27. Moghadamtousi SZ, Fadaeinasab M, Nikzad S, Mohan G, Ali HM, Kadir HA. Annona muricata (Annonaceae): a review of its traditional uses, isolated acetogenins and biological activities. International journal of molecular sciences. 2015 Jul 10;16(7):15625-58.
28. Tambun R, Alexander V, Ginting Y. Performance comparison of maceration method, soxhletation method, and microwave-assisted extraction in extracting active compounds from soursop leaves (Annona muricata): A review. InIOP Conference Series: Materials Science and Engineering 2021 Mar 1 (Vol. 1122, No. 1, p. 012095). IOP Publishing.
29. Nguyen MT, Nguyen VT, Minh LV, Trieu LH, Cang MH, Bui LB, Le XT, Danh VT. Determination of the phytochemical screening, total polyphenols, flavonoids content, and antioxidant activity of soursop leaves (Annona muricata Linn.). InIOP Conference Series: Materials Science and Engineering 2020 (Vol. 736, No. 6, p. 062011). IOP Publishing.
30. Aguilar-Hernández G, Zepeda-Vallejo LG, García-Magaña MD, Vivar-Vera MD, Pérez- Larios A, Girón-Pérez MI, Coria-Tellez AV, Rodríguez-Aguayo C, Montalvo-González E. Extraction of alkaloids using ultrasound from pulp and by-products of soursop fruit (Annona muricata L.). Applied Sciences. 2020 Jul 16;10(14):4869.
31. Vinothini R, Growther L. Isolation and identification of acetogenin from Annona muricata leaves. Int. J. Appl. Biol. Pharm. Technol. 2016;7:169-72.
32. Gajalakshmi S, Vijayalakshmi S, Devi RV. Phytochemical and pharmacological properties of Annona muricata: a review. International Journal of Pharmacy and Pharmaceutical Sciences. 2012 Oct;4(2):3-6.
33. Abubakar M, Koul B, Chandrashekar K, Raut A, Yadav D. Whitefly (Bemisia tabaci) management (WFM) strategies for sustainable agriculture: a review. Agriculture. 2022 Aug 26;12(9):1317.
34. de Silva WI, Haroon MH. Insecticidal activity of soursop leaves (Annona muricata L.) against Sitophilus zeamais.
35. Goud BR, Raghavendra M, Prasad PS, Hatti V, Halli HM, Nayaka GV, Suresh G, Maheshwari KS, Adilakshmi G, Reddy GP, Rajpoot SK. Sustainable management and restoration of the fertility of damaged soils. Agriculture Issues and Policies. 2022;113.
36. Perring TM, Stansly PA, Liu TX, Smith HA, Andreason SA. Whiteflies: Biology, ecology, and management. InSustainable management of arthropod pests of tomato 2018.
37. Kawalekar JS. Role of biofertilizers and biopesticides for sustainable agriculture. J Bio Innov. 2013 Sep 11;2(3):73-8.
38. Boica AL, Macedo MA, Chagas Filho NR, Angelini MR, Boiça Neto AL. Control strategies for Bemisia tabaci (Genn.) biotype B and other insect vectors in tomato lycopersicum esculentum (Mill.).
39. Ragasa CY, Galian RF, Shen CC. Chemical constituents of *Annona muricata*. Der Pharma Chemica. 2014;6(6):382-7.

40. Wahua C, Ogan I. Micro-Morphological and Proximate Analysis of Soursop (Annona muricata L. of Annonaceae) Harvested from a Tertiary Institution Campus in the Niger Delta Region Nigeria. Journal of Applied Sciences and Environmental Management. 2024 Jan 29;28(1):85-9.
41. Ummu Hani B, Nur Munirah S, Tan AL. MICROSCOPIC CHARACTERISTICS OF *Annona muricata L.*(ANNONACEAE) FOR AUTHENTICATION OF HERBAL RAW MATERIAL. Unravelling Nature's Treasures & Secrets: Current Species of Interest.:228.24 510–513.
42. Zhao G, Hui Y, Rupprecht JK, Mclaughlin JL. Assay of annonaceous acetogenins by HPLC with an evaporative light- scattering detector. J Nat Prod.1992;55(5):347-56.
43. Kumar D, Kumar A, Prakash O, Rathi A. Determination of ash value, moisture content and extractive values of root of Ficus hispida. Asian J Pharm Clin Res.2012;5(2):168-71
44. Mulia K, Krisanti E, Maulana T. SELECTIVE POLARITY-GUIDED EXTRACTION AND PURIFICATION OF ACETOGENINS IN Annona muricata L. LEAVES. International Journal of Technology. 2015 Nov 1;6(7).
45. Luna, J.D.S., Carvalho, J.M.D., Lima, M.R.F.D, Bieber, L.W., Bento, E.D.S., Franck, X., Santana, A.E.G., (2006).
46. Acetogenins in *Annona muricata L.* (annonaceae) Leaves are Potent Molluscicides. Natural Product Research, Volume 20(3), pp. 253-257
47. Tuzimski T, Sherma J In: Meyers RA (ed) Thin layer chromatography, vol15. Wiley, Chichester, UK 2000:1-26.
48. Pereira ID, Vega MR. Flavonoids in the Annona genus: Chromatographic and Spectral Methods of Analysis–A Review.
49. Moghadamtousi SZ, Fadaeinasab M, Nikzad S, Mohan G, Ali HM, Kadir HA. Annona muricata (Annonaceae): a review of its traditional uses, isolated acetogenins and biological activities. International journal of molecular sciences. 2015 Jul 10;16(7):15625-58.
50. Oyedeji O, Taiwo FO, Ajayi OS, Ayinde F, Oziegbe M, Oseghare CO. Biocidal and phytochemical analysis of leaf extracts of Annona muricata (Linn.). Int J Sci Basic Appl Res. 2015;24(7):76-87.
51. Yang H, Li X, Tang Y, Zhang N, Chen J, et al. (2009) Supercritical fluid CO2 extraction and simultaneous determination of eight annonaceous acetogenins in annona genus plant seeds by HPLC-DAD method. J Pharm Biomed Anal 49: 140-144
52. Errayes AO, Abdussalam-Mohammed W, Darwish MO. Review of phytochemical and medical applications of *Annona muricata* Fruits. J. Chem. Rev. 2020;2:70-9.
53. De Barro PJ, Liu SS, Boykin LM, Dinsdale AP, Bemisia tabaci: A statement or species status. Annu Rev Entomol.2011;56:1-19.
54. Nguyen MT, Nguyen VT, Minh LV, Trieu LH, Cang MH, Bui LB, Le XT, Danh VT. Determination of the phytochemical screening, total polyphenols, flavonoids content, and antioxidant activity of soursop leaves (Annona muricata Linn.). InIOP
55. Gupta A, Rao S, Dubey A. Seed morphology of Annona muricata Linn. Journal of Seed Research.

2018;2(1):1-4.

56. Zhao G, Hui Y, Rupprecht JK, Mclaughlin JL. Assay of annonaceous acetogenins by HPLC with an evaporative light- scattering detector. J Nat Prod.1992;55(5):347-56.
57. Kim, G.S., Zeng, L., Alali, F., Rogers, L.L., Wu, F.E., Sastrodihardjo, S., McLaughlin, J.L., (1998). Muricoreacin and Murihexocin C, Mono-tetrahydrofuran acetogenins, from the Leaves of *Annona muricata*. Phytochemistry, Volume 49(2), pp. 565-571
58. Moghadamtousi, S.Z.; Fadaeinasab, M.; Nikzad, S.; Mohan, G.; Ali, H.M.; Kadir, H.A. Annona muricata (Annonaceae): A Review
59. of Its Traditional Uses, Isolated Acetogenins and Biological Activities. Int. J. Mol. Sci. 2015, 16, 15625–15658.

60. F.E. Wu, L. Zeng, Z.M. Gu, G.X. Zhao, Y. Zhang, J.T. Schwedler, J.L. McLaughlin. J. Nat. Prod., 58, 902 (1995)